

WHAT IS CLAIMED IS:

- 09610935-070600
1. A composition suitable for formulation of an enzymatic reaction mixture, the composition comprising a reaction component essential for an ex-vivo non-
5 polymerase enzymatic reaction in which a substrate is catalyzed by an enzyme in a reaction mixture to form a product, and a tracer compatible with the enzyme, the composition being substantially free of the substrate.
 2. The composition of claim 1 having a density of at least about 1.01 g/cm³.
 3. The composition of claim 1 having a density of at least about 1.1 g/cm³.
 4. The composition of claim 1 having an optical density greater than about 5 at a visible wavelength of maximal tracer absorbance.
 5. The composition of claim 1 wherein the optical density of the composition is at least about 15 at a visible wavelength of maximal tracer absorbance.
 6. The composition of claim 1 wherein the optical density of the composition is about 200 - 400 at a visible wavelength of maximal tracer absorbance.
 7. The composition of claim 1 wherein the reaction component essential for an enzymatic reaction comprises a concentrated buffer solution.
 8. The composition of claim 1 wherein the reaction component essential for an enzymatic reaction comprises an enzyme.

9. The composition of claim 8 wherein the enzyme is a nucleic acid-modifying enzyme.

10. The composition of claim 1 wherein the tracer is comprised of a dye selected from the group consisting of acid violet 5, acid red 1, and amaranth.

Sub C.7
11. A composition comprising a reaction component essential for an ex-vivo enzymatic reaction in which a substrate is catalyzed by an enzyme in a reaction mixture to form a product, and a tracer compatible with the enzyme, the composition being substantially free of the substrate and having an optical density greater than about 5 at a visible wavelength of maximal tracer absorbance.

Q2
12. The composition of claim 11 wherein the reaction component is essential for an ex-vivo polymerase reaction in which a nucleic acid polymer product complementary to a nucleic acid polymer template is prepared, the tracer is compatible with the polymerase, and composition is substantially free of the nucleic acid polymer template.

Sub C.7
13. The composition of claim 12 wherein the density of the composition is at least about 1.01 g/cm^3 .

14. The composition of claim 12 wherein the density of the composition is at least about 1.1 g/cm^3 .

15. The composition of claim 12 wherein the optical density of the composition is at least about 15 at a visible wavelength of maximal tracer absorbance.

009020 SE601960

16. The composition of claim 12 wherein the optical density of the composition is about 200 - 400 at a visible wavelength of maximal tracer absorbance.

17. The composition of claim 12 wherein the reaction component essential for a polymerase reaction is a concentrated buffer solution.

18. The composition of claim 12 wherein the reaction component essential for a polymerase reaction is a polymerase.

19. The composition of claim 18 wherein the polymerase is a thermostable polymerase.

Sub 237 20. The composition of claim 19 wherein the polymerase is Taq polymerase.

21. The composition of claim 12 wherein the tracer is comprised of acid violet 5 and acid red 1.

22. The composition of claim 14 wherein the optical density of the composition is about 200 - 400 at a visible wavelength of maximal tracer absorbance, the reaction component essential for a polymerase reaction is a Taq polymerase, and the tracer consists of 20% acid violet 5 and 80% acid red 1.

23. In a method for a polymerase reaction that comprises

(a) forming a reaction mixture comprising a polymerase, a nucleic acid polymer template, a tracer compatible with the polymerase, and other components essential for the polymerase reaction,

- (b) creating a nucleic acid polymer product complementary to the nucleic acid polymer template by enzymatic reaction,
- 10 (c) analyzing the product of the enzymatic reaction by an electrophoretic protocol, and
- (d) observing the tracer during the electrophoretic protocol without providing additional tracer beyond that which was included in the reaction mixture, the
- 15 improvement comprising

supplying the tracer to the reaction mixture in a composition that comprises the tracer and the enzyme or another essential component, the composition being substantially free of the nucleic acid polymer template.

24. In the method of claim 23, the improvement further comprising the reaction mixture having an optical density at least about 15 at a visible wavelength of maximal tracer absorbance.

25. In the method of claim 23, the improvement further comprising the reaction mixture having a density at least about 1.01 g/cm³.

26. In the method of claim 23, the improvement further comprising the tracer consisting of a combination of acid violet 5 and acid red 1.

27. In the method of claim 25, the improvement further comprising the reaction mixture having an optical density at least about 15 at a visible wavelength of maximal tracer absorbance, and the tracer consisting of a
5 combination of 20% acid violet 5 and 80% acid red 1.

28. A method for a restriction enzyme reaction, the method comprising forming a reaction mixture comprising a

009020" 52607360

5 restriction enzyme, a nucleic acid polymer substrate, a
tracer compatible with the restriction enzyme, and other
components essential for the enzymatic reaction,
enzymatically cleaving the nucleic acid polymer substrate
to form a restriction product, analyzing the restriction
product by an electrophoretic protocol, and observing the
tracer during the electrophoretic protocol without
10 providing additional tracer beyond that which was
included in the reaction mixture.

29. The method of claim 28, wherein the optical
density of the reaction mixture is at least about 15 at a
visible wavelength of maximal tracer absorbance.

30. The method of claim 28, wherein the density of
the reaction mixture is at least about 0.01 g/cm greater
than the liquid phase utilized in the chromatographic or
electrophoretic protocol.

31. The method of claim 28, wherein the tracer is
amaranth dye.

32. The method of claim 29, wherein the density of
the reaction mixture is at least about 0.01 g/cm greater
than the liquid phase utilized in the chromatographic or
electrophoretic protocol, and the tracer is amaranth dye.

33. The method of claim 28 wherein the tracer in
the reaction mixture is of adequate character and
sufficient quantity to be visible during the
electrophoretic protocol.

09610935 070600

Call AB